1 INTRODUCTION

1.1 Melanoma

Cutaneous melanoma has one of the most rapidly increasing incidence rates of any cancer in the U.S. Early detection and surgical excision of melanoma can be curative, but the outlook for patients with melanoma that has spread beyond the primary is poor. Currently, there are no clearly effective therapies for advanced (stage III and IV) disease. Such patients can be expected to have a median survival ranging from 8.1 (stage IV) to 32 months (stage III) (l-4).

Adjuvant interferon-alpha-2-b initially demonstrated a survival advantage in patients with thick (> 4mm) primaries or regional nodal involvement (5). Unfortunately, a follow-up study failed to confirm a survival benefit with this same regimen (6). As a result, the benefit of adjuvant interferon for patients with advanced melanoma has been drawn into question (3).

Dimethyl triazeno imidazole carboxamide (dacarbazine or DTIC) is currently the only chemotherapeutic with an indication in melanoma. Response rates for DTIC in stage IV disease range from 10.1 to 12.1% in two recent randomized trials (7,8). Multiple drug regimens have been evaluated that appear to offer increased response rates at the expense of increased toxicity and no clear survival advantage compared to DTIC (8,9).

1.2 Immune System and Cancer

The role of the immune system in malignant melanoma has been an area of intense interest. It has been documented that patients with progressive tumor growth have impaired immune function (10,11). Agents that can stimulate non-HLA-restricted cellular cytotoxicity, such as interferon and interleukin-2, have produced sustained regressions in some patients. However, the toxic side effects of these agents are considerable, with several deaths reported following the use of high-dose regimens (3,4,12,13).

Attempts have also been made to generate tumor specific (HLA-restricted) immune responses. Recognition of foreign tumor antigens by the immune system requires presentation of antigen peptide fragments in the context of MHC-class I or class II molecules (14,15). $CD8^+$ cytotoxic T lymphocytes (CTLs) are activated by MHC-class I bearing cells, while $CD4^+$ T-cells are stimulated primarily by tumor peptides presented by cell surface MHC-class II molecules. MHC-class I levels in tumor cells, including melanoma, are often decreased or undetectable. This may represent one mechanism by which tumor cells escape immune rejection (16,17). In some cases, a correlation has been demonstrated between HLA loss and poor prognosis (18-20).

It is possible to re-introduce MHC-class I expression in tumors via DNA-based therapy. Allovectin-7® has been developed for this purpose. Allovectin-7® consists of a DNA plasmid (VCL-1005) containing the genes for an allogeneic MHC-class I protein, human leukocyte antigen B7 (HLA-B7), and β2-microglobulin complexed with a cationic lipid mixture (DMRIE/DOPE) that aids in the uptake of VCL-1005 into tumor cells. HLA-B7 was chosen because of its infrequent expression in the U.S. population and the additional possibility of developing an allogeneic immune response in HLA-B7 negative patients (2 1,22). β2-microglobulin was included in VCL-1005 in order to allow for the expression of the complete MHC complex on the cell surface. VCL-1005 provides several potential immune-stimulating functions: a foreign cell surface protein (i.e. HLA-B7 in HLA-B7 negative patients), T-cell

activation, and an antigen-presenting activity signal (antigen presented in the context of HLA-B7) (23).